In its application to blood the use of the name is unnecessary, since the only "buffer-salt" of any importance is the sodium bicarbonate of the plasma; phosphate is present here only in insignificant amount. In its behaviour towards acids the plasma reacts precisely like solutions of sodium bicarbonate, and so little doubt is there as to the existence of this salt in it that until recently the carriage of the carbon dioxide from the tissues to the lungs was generally believed to be effected by it. It is truly present in less quantity by weight than the proteins, but quite sufficient to neutralize the fixed acids produced in the tissues. The molar concentration, indeed, of bicarbonate is higher than that of protein on account of the enormous molecular weight of the latter. In the opinion of Dr. Moore the function of combining with excess acid is to be ascribed to the proteins, not to the bicarbonate. He gives no data for the statement that there are not enough salts to account for one-tenth of the "protective influence." But I take it that he refers to his titrations with strong acid and alkali, to be discussed below. As to the carriage of carbon dioxide, he appears to be unaware of the work of Buckmaster, who has shown that this function belongs to haemoglobin.

The proteins of the plasma are said to be responsible for at least 90 per cent. of the "protective action" of the blood. I confess that I cannot understand the meaning of the statement that "all the inorganic salts of the plasma, including even the sodium chloride, are held in union by the proteins." How, then, can the fact be explained that the electrical conductivity of the plasma is practically that of a solution of free salts? It is also inconsistent with what is pointed out in the next paragraph, namely, that the depression of the freezing point is that of a one-sixth normal sodium chloride; in other words, about that of the salts, if free. If the salts were combined with the proteins, the depression of the freezing point would be much less than this. It is true that Pfeiffer and Modelski¹ have described the formation of what they regard as chemical compounds between certain amino acids and certain neutral salts; but it is remarkable that sodium chloride was not one of the salts, and I have been unable to confirm their results, although the instructions given were followed exactly. Much less is there evidence of combination between proteins and salts. On the contrary, the freezing-point measurements of Bugarsky and Liebermann² showed distinctly that this was not the case. Hardy³ also was unable to find any evidence of chemical combination between salts and globulins.

The reason why Moore and his coadjutors found serum proteins to be able to neutralize acids and alkalis was because they used *strong* acid and alkali. Such are never sent into the blood stream as products of metabolism, and the representation of the "buffer" action of blood as related to these unphysiological reagents is incorrect. There is no evidence that proteins combine with the relatively weak organic acids produced in tissue metabolism. If an indicator such as neutral red, which reacts to small changes of hydrogen ion just around the point of neutrality, be added to serum, it will be found to change colour when a very small amount of lactic acid is put in, showing that the proteins have not taken it up. The change of colour can easily be shown to be due to the carbon dioxide liberated from the bicarbonate, since it can be brought back on exposure to air. The production of the carbamino acids of Siegfried is limited to the simpler amino acids; according to this observer himself, even leucine does not form one. However, I found, by conductivity measurements signs of combination of carbon dioxide with leucine. But there is no trace of the effect with proteins. We may also call to mind the results of Fletcher and Brown,⁴ who showed that the only carbon dioxide given off by muscle on heating arises from the decomposition of sodium bicarbonate by the lactic acid formed. This applies up to the temperature at which destruction of the proteins occurs. Before the view put forward by Moore can be accepted, it is necessary that experimental evidence be brought showing that a pure protein solution can take up in combination sufficient carbon dioxide to account for that given off by plasma on the addition of a strong acid. Findlay⁵ shows that no carbon dioxide is taken up by egg white, a very small amount by gelatin.

But there is no doubt that proteins are able to form true salts with strong acids and bases. They can combine with the former by aid of their basic (NH_2) groups and

with the latter by aid of their acidic (carboxyl) groups. The reason why they do not combine with weak acids or neutral salts is clearly that the NH_2 and COOH groups are, in neutral solutions, united together by anhydride formation. In order that any combination may take place with other substances, this internal anhydride structure must be broken down by hydrolysis, a process that requires the aid of a strong acid or base.

It is somewhat remarkable that serum proteins are, chemically, very inactive substances. They do not serve for nutrition of the tissues, and their chief function seems to be to give a colloidal osmotic pressure to the blood, so that it shall not lose water to the tissues too rapidly. They have also an important part to play in the clotting process. I venture to think, however, that the term "reactivity of the serum" is not a particularly lucid one. It is usually associated with the capacity of readily entering into chemical reaction with other substances.

The fact that salts are formed by proteins with strong acids and bases is the cause of the rise of osmotic pressure observed in such cases by Roaf and Adamson. These salts are dissociated electrolytically, and, as I have shown elsewhere, all the ions are osmotically active. Various explanations might be given of the fact referred to by Moore, that the addition of a very small amount of acid lowers the osmotic pressure. It may be due to the decomposition of a small quantity of a sodium salt of protein present in the serum owing to escape of the normal carbon dioxide and the consequent development of alkalinity.

The use of the name "buffer-salts" in application to blood plasma is to be deprecated, as it seems to me, because it is apt to give an air of mystery to what is merely a simple property of bicarbonates. The expression "sodium bicarbonate" is all that is wanted in place of "buffer-salts" in this connexion.

No satisfactory name has been suggested for the general property as possessed by many weakly dissociated electrolytes. "Tampon" is better than "buffer" in any case.

REFERENCES.

- ¹ Hoppe-Seyler's *Zeitsch.*, vol. lxxxi, p. 329; and vol. lxxxv, p. 1.
² *Pflüger's Archiv*, vol. lxxii, p. 51. ³ *Journ. of Physiol.*, vol. xxxiii, p. 251. ⁴ *Ibid.*, vol. xlviii, p. 177. ⁵ *Kolloid. Zeitsch.*, vol. iii, p. 169.

THE PART PLAYED BY THE BONE GRAFT.

BY

MARCUS MAMOURIAN, F.R.C.S.E.,

SURGEON TO THE DISTRICT INFIRMARY AND CHILDREN'S HOSPITAL, ASHTON-UNDER-LYNE.

THE subject of bone grafting, which has acquired so great an importance to-day, has engaged the attention of surgeons for more than a century. Albee mentions Merren as having carried out experiments in 1809. The wonderful researches and operations of Ollier and Macewen appeared as long ago as 1867 and 1881. Singularly enough, the biology of bone and the bone graft still remains undetermined. The main views may be summarized as follows:

1. Bone is osteogenetic. The bone graft is osteogenetic and can live and grow without periosteum. (Macewen, Groves.)
2. Fragments of bone grow better than an entire piece. (Macewen.)
3. Periosteum acts as a limiting membrane and has no osteogenetic power. (Macewen.)
4. The periosteum is the principal agent of bone growth. The bone graft can live and grow by virtue of its own periosteum. (Ollier, Axhausen, and most of the American writers.)
5. The bone graft is osteo-conductive. (Murphy.)
6. Bier and others attach great importance to the cambium layer, endosteum, Haversian canal linings and marrow.

1, 2, 3. Macewen is the protagonist of the "bone from bone" theorists. His teaching is mainly founded upon his famous case (1882) in which part of the humerus of a boy was excised for osteomyelitis and restored by the implantation of small sections of bone obtained in the course of several osteotomies. The graft, therefore, was comminuted and homoplastic, and the logical inference is that the reconstruction of the diaphysis was brought about by the increase in size and coalescence of the fragments of bone. In the absence of a radiographic record all explanation as to the process of restitution must be more or less conjectural, or, at most, based upon experimental findings. The remarkable feature of the case is, that although the shaft is supposed to have been restored without the help of a "limiting membrane," yet there was limitation of size

and shape. Now, this is explained by "the law of natural adaptation," which seems to be the basis of all the theories on the survival and growth of bone grafts. It may be pertinently asked, however, if this law is potent enough to enable a parasitic piece of bone to acquire vitality, growth, and shape, how is it that, often enough, it fails to influence the two segments of a broken bone even when placed by open operation in ideal apposition and alignment? Macewen's technique is a direct challenge to the present practice of utilizing autogenous bone grafts covered with periosteum and firmly contacted with the recipient bone ends.

4. The teaching of the periosteum school is based upon the histological evidence that the cells of the periosteum of the bone graft show active proliferation. The loss of nuclear staining and the ultimate absorption of the bone cells is generally admitted, hence the convenient hypothesis that the vascularized strip of periosteum is the parent of the new bone. McWilliams is so strong in this belief as to doubt the possibility of successful bone grafting if the graft be denuded of its periosteum; while Groves is equally emphatic in his contention "that the process takes place by the proliferation of the cells of the bone itself." Macewen's work is sufficient refutation of McWilliams's claim, and my case J. A., with a complete x-ray record, is additional proof that bone can regenerate by means of grafts (I use the phrase advisedly) without periosteum. If the periosteum is the bone-forming medium, why advocate "whole thickness" grafting with rigid fixation? why adopt a procedure which can only be justified by the assumption that the graft can live in its entirety?

5. Murphy considered the problem on Barth's lines, and advanced the view that the graft is osteo-conductive—that is, that it acts as a scaffolding and gets gradually absorbed. He thus differs from the rest in admitting neither periosteal osteogenesis nor the survival of osteoblasts.

In the radiograms of my cases, taken at frequent intervals during the last four years, the disappearance of the graft can be traced in the clearest possible manner, and in no instance have I been able to verify "the increase in thickness which is considered to be the single sure sign of the continuance of the life of the graft" (Ollier, McWilliams). The increase in thickness is due to the deposit of new bone around the graft and not to any hyperplasia of the graft itself.

The great diversity of opinion amongst experimentalists leads us to the true conclusion, which is, that all the elements that enter into the constitution of bone are required for osteogenesis, and in agreeing that the best results are obtained from autogenous bone grafts with their periosteum and endosteum, all practical surgeons confirm this opinion.

It has generally been taken for granted, however, that the bone graft, once vascularized, behaves like normal bone, notwithstanding the fact that bone implanted anywhere but in a bone space always gets absorbed. No one has ever been able to produce progressive bone growth in a region where there has been no bone before.

In all these polemics there are two factors which do not appear to have received adequate consideration—namely, the periosteum and bone of the grafted limb. In no case could it be assumed that the shaft and periosteum have been destroyed or removed in their entirety by disease or excision. As a rule portions of the diaphysis and the whole periosteal sheath remain, and, even under the worst conditions, shreds of periosteum and spicules of bone are left in the gap.

What is, then, the part played by the bone graft from the time of its insertion to its disappearance?

In my opinion, all that the graft does is to supply what may be called the biochemical stimulus or irritant which has been abolished by trauma or disease, the new bone being formed from the diaphyseal ends, from periosteal and bony remains in the shaft zone, and in the young by epiphyseal lengthening. Dead bone acts in a similar way, but much less effectively. This is the explanation of Macewen's case and of my case J. A., with practically identical features. How else is it possible to account for the restoration of bone even after the graft has been actually excluded like a sequestrum and the graft bed in a state of profuse suppuration? The cambium layer and Haversian linings are cited by some as explaining these occurrences. No cambium or any other layer can float in pus and live.

The stimulating property of the bone graft has been noted by many, but only in the sense of a limited degree of cellular activity sufficient to cause fusion between the graft and the diaphyseal stumps. No one, however, seems to have considered this stimulus as the essential and all-important function of the graft.

Distinction must also be made between the use of a graft in an inert and in an active osteogenetic field. In cases of excision of the diaphysis in the young the new bone is often attributed to the graft, though regeneration usually takes place whether a graft is used or not. But even in children the graft is indicated, because it serves as an intraperiosteal splint, stimulates and accelerates bone growth, with the result that the shaft is reproduced perfectly and in less time.

Albee's book is full of illustrations intended to show the increase in size of the graft and the evolution of the medullary cavity, but in most of them the outline of the periosteum is discernible owing to the laminae of bone still adhering to it.

The success of the inlay method is due to the facts that it entails the exposure of the healthier sections of the bones and that each tissue is stimulated by its like. The more anatomical the "sitting" of the graft the better the result. When a graft is not absorbed and remains identifiable for six months or more, even though seemingly united by callus, the union will be found to be false and fragile and the graft undergoing cartilaginous change. (Case T. E.)

Bone graft surgery is not as widely practised as it deserves to be. In the mutilations of war, osteomyelitis, tuberculous disease, ununited fractures—in short, in all cases necessitating excision of bone, in spinal caries and in fractures of the spinal column the bone graft is an unrivalled means of repair and cure.

DESCRIPTION OF CASES.

Case 1.

J. A., aged 8, operated on for acute osteomyelitis, August, 1913, came under my care in August, 1914. There had not been the slightest attempt at bone growth for twelve months. The gap measured over six inches; the diaphyseal ends were nipple-shaped and sclerosed. An autogenous bone graft without periosteum was placed *in situ*; its upper end was placed in "shavings" raised with the chisel, and the lower end in the medullary canal (Fig. 1). There was no firm contact in Murphy's sense. The graft was cut with chisels. Fig. 2 (September 1st, 1916) shows great increase in length of the upper segment. The shaft was still incomplete, the gap measuring about 1½ in. with the rib in position. The parents objected to more bone being taken from the sound leg. Rarefaction of the lower segment was noticeable in the radiogram. The rib was completely absorbed. Fig. 3 shows inlay grafting without periosteum (circular twin saw) in February, 1917, and Fig. 4 the completed shaft.

Case 2.

A. F., aged 9, had tuberculous diaphysitis of the radius of many years' standing; there was mixed infection, with profuse suppuration and sinuses. Fig. 5 shows the extensive involvement of the shaft.

Excision of diaphysis was performed and graft without periosteum inserted, as shown in Fig. 6. Small drainage tubes are seen in the periosteal sheath and laminae of bone adhering to periosteum. Fig. 7 shows active osteogenesis and the shapelessness of the new bone. The graft is faintly visible. Fig. 8, from a radiogram taken after eight months, affords evidence of the perfect restoration of the shaft.

Case 3.

H. B., aged 17, was suffering from tuberculous disease of a phalanx (Fig. 9). A graft without periosteum was placed in the periosteal sheath (Fig. 10). Bone formation around the graft is seen in Fig. 11, and absorption of the graft and completion of the phalanx in Fig. 12.

Case 4.

T. E., aged 16, was seen with compound fracture of the tibia and virulent sepsis. Fig. 13 shows the loss of bone and the nipple-shaped and sclerosed diaphyseal ends. There was no sign of new bone for twelve months. Inlay grafting with periosteum (circular twin saw) was performed in December, 1915. By May, 1916, there was firm union. Fig. 14, from a radiogram taken on readmission to hospital for fracture in the region of graft in August, 1917, shows the graft still identifiable, and the callus above and below and the fracture. At the operation the graft was found to be quite soft and was cut out with the knife. The part was regrafted (inlay with periosteum) and there is now firm union.

I am indebted to Dr. Woodburn Morison, radiographer to the District Infirmary, for the radiograms.

Owing to the difficulty of reproducing radiograms under existing conditions, due to shortage of paper, drawings in Indian ink, made from the radiograms by Mrs. Williamson, of Ashton-under-Lyne, have been reproduced here.

J.A. 1
FIG. 1.

J.A. 2.
FIG. 2.

J.A. 3.
FIG. 3.

J.A. 4
FIG. 4.

A.F. 1,
FIG. 5.

A.F. 2.
FIG. 6.

A.F. 3.
FIG. 7.

A.F. 4
FIG. 8.

H.B. 1
FIG. 9.

H.B. 2
FIG. 10.

H.B. 3
FIG. 11.

H.B. 4
FIG. 12.

T.E. 1.
FIG. 13.

T.E. 2
FIG. 14.